REMARKS

1. Objections and 112/2 Issues

1.1. Claim 78 has been made dependent on 77, as intended.

1.2. Claim 60 was rejected under 112/2 because there was a lack of antecedent basis for "the initial cytotoxic T cells" and "the tissue sample". We have excised the phrase which included these recitations, and instead state that the functional profile "is not substantially altered during continuous growth". For basis, see page 10, lines 3-5.

1.3. In claim 61, the recitation of "normal" was inadvertent; just "disease-associated" was intended. Hence, "normal" has been deleted.

1.4. Claim 63 is amended to delete "functionally similar" and add IL-12, to overcome a lack of clarity objection. In response to the examiner's question regarding the properties of said cytokines, we confirm that all the cytokines of claim 63 promote cytotoxic T cell growth, which by definition is an activating property.

1.5. Claim 69 is amended to delete the term "body fluid", since bone marrow is defined as a soft tissue (see Cruse et al P39).

1.6. Claim 71 is amended to delete the term "or cells having similar properties".

1.7. Claim 72 is amended to delete the term "and functionally similar compounds" and to place it in correct Markush form.

1.8. Claim 75-79 are amended to claim "a pharmaceutical composition" in place of "an immunological composition". Claim 75 is further amended to be directed to a pharmaceutical composition comprising disease associated antigen activated cytotoxic T-cells according to claim 40 and one or more excipients selected from the group consisting of water, saline,

dextrose, glycerol, and ethanol. Because of the "one or more" language, it covers use of combinations of the recited excipients.

2. Description (112/1) Issue

The Examiner is of the opinion that "immunological compositions" is "new matter", but concedes disclosure of both the broader concept "pharmaceutical compositions" and of the narrower concept "vaccine compositions".

Claims 75-79 have been amended to refer to pharmaceutical compositions.

3. Prior Art Issues

The following prior art rejections have been maintained or added:

(1) claims 40, 60, 70-71, 73, 75-77 and 85-88 as anticipated by Riddell (5,827,642) under 102(e)

(2) claims 40, 60-73, 75-77 and 85-88 as anticipated by Haberman (5,188,959) under "103(b)" [sic, "102(b)"]

(3) claims 40, 60-65, 68-73, 75-77 and 84-88 as anticipated by Flyer (6,316,257) under 102(e)

(4) claims 40, 60 and 73-74 as obvious over Riddell, Haberman or Flyer in view of Boll et al.

(5) claims 40, 75 and 79 as obvious over Riddell, Haberman or Flyer in view of Santoli (5,702,702).

Claim 40, as examined, recited:

> A disease associated, antigen activated cytotoxic T-cell line, wherein the cells have exceeded or have an expected life-span of at least 40 PD.

Dependent claim 84 recited,

> A cell line according to claim 40 wherein the cells have exceeded 40 PD.

Note that only rejection (3) above was applied to claim 84.

The actual lifespan limitation of claim 84 has been imported into amended claim 40, and hence 84 has been cancelled as redundant.

3.1. Anticipation by Riddell

We do not need to consider at this time whether the cells of Riddell would reasonably have been expected to have an expected lifespan of 40PD, as amended claim 40 requires that the cells have in fact expanded so far, i.e., that they actually have exceeded 40 PD.

Claim 84, reciting an actual lifespan limitation, was not rejected as anticipated by Riddell, which would appear to imply that the amendment of 40 to incorporate the limitation of 84 moots this rejection.

However, in the paragraph bridging pp. 5-6 of the office action, the Examiner suggests that Riddell achieved an actual lifespan of 39-42 PD:

> Furthermore, the expansions taught by Riddell et al are not representative of the total degree of expansion (number of generations or population doublings) that the T-cells have undergone. Prior to subjecting T-cells to their taught "rapid expansion method" (REM), the cells have already undergone a number of divisions. That is, what Riddell et al teach is a method that is to be conducted on an already established T-cell clone (col. 9, lines 43+) which itself has previously been dividing (e.g. col. 10, lines 45+). Thus all of applicant's calculations set forth in the traverse of the rejection do not reflect the actual number of divisions/doublings that the T-cells have undergone. One would, in fact, need to add to the number of clonal cell divisions (col. 9, lines 62-64), which applicant calculates as 30 PD, the number of REM cell divisions (col. 9 lines 38-40), which applicant calculates as 9-12 PD. The

examiner thus calculates the total PD as 39-42.

The Examiner has misunderstood the calculations presented on page 8 of the last amendment. Our understanding of Riddell col. 9 was that the total expansion which he had achieved was to 10^9 cells, equivalent to 30 PD, and that this was achieved by applying his REM to cells which were already expanded.

Thus, in Example 1, the input cell value was 5-6x10^4 T cells (col. 18, lines 22-24), equivalent to a PD of 16, and REM was used to expand these cells 866- to 1500-fold (col. 18, lines 54-55), i.e., another 11 PD, for a total PD of 27. The final count, shown in Fig. 1, was at most 7.5 x 10^7 cells.

Likewise, in Example 2, the input cell number was 10^5-5x10^5 cells (col. 18, line 67 to col. 19, line 1), and these cells were expanded 250- to 2200-fold (col. 19, line 18) by the REM method to obtain a population of 6.2x10^8 to 9x10^9 cells, see col. 19, line 20 and Fig. 2, the latter being equivalent to a PD of 33-34.

The Examiner's error was thus that of adding, to the total PD of 30 (10^9 cells), an expansion value for a single step already included in that total PD.

3.2. Anticipation by Habermann

The Examiner's defense of the Haberman anticipation rejection emphasizes that the rejected claims merely require an expected lifespan of 40 PD. Claim 84, reciting actual accomplishment of 40 PD, was not so rejected, implying that amended 40 distinguishes Haberman. Haberman only achieved an expansion equivalent to 23-26 PD.

3.3. Anticipation by Flyer

Similarly, the examiner finds the claims 40, 60-65, 68-73, 75-77 and 84-88 lack novelty over Flyer (US 6,316,257). Flyer

teaches a modified REM method for the expansion of cytolytic T-cells. Example 1 describes a pre-MREM population of $5x10^4$ CTL expanded to a final population of $4.86x10^7$. Starting from a single progenitor cell, this final population corresponds to a total PD of 26. Considering all the examples, the maximum population attained by culturing was less than 10^8, corresponding to a PD of less than 27.

While the examiner refers to a "mammalian cell line" which according to Flyer may be propagated in vitro for "at least about 100 generations", there is no teaching that these mammalian cells are T cells.

3.4. Obviousness Rejections

The examiner rejects claims 40, 60, and 73-74 as obvious in view of Riddell, Haberman, or Flyer in combination with Boel, which describes use of cytotoxic T cells for treatment of melanoma. Similarly the examiner rejects claims 40, 75, and 79 as obvious in view of Riddell, Haberman, or Flyer in combination with Santoli, which describes expansion of T cell lines with cytokines, and irradiating the cells to prevent proliferation prior to in vivo administration. None of the cited primary references disclose cytotoxic T cells having a life span of at least 40 PD.

Riddell asserts at col. 19, lines 22-27 that

> Repetitive cycles of stimulation are also effective in promoting rapid expansion; thus, with this technique it is possible to generate greater than 10^{15} clonally-derived effector T cells. In practice, cell doses of 10^9 to $5x10^{10}$ are more feasible (due to the media and incubator space required for very large scale cultures) and are generally sufficient for use in adoptive immunotherapy.

The Examiner will note that this passage uses the present

tense, implying that Riddell had not in fact attempted to obtain cell populations higher than $9x10^9$ cells by additional REM cycles. The question, then, is whether it would be reasonable to expect that by such additional REM cycles, populations exceeding 40 PD (claim 40) or even achieving at least 50 PD (claim 89) or 60 PD (claim 90) would be obtained by a person of ordinary skill in the art. We think not.

In Riddell Example 1, $25x10^6$ PBMC feeder cells and $5x10^6$ LCL feeder cells were used to stimulate the growth of $5\text{-}6x10^4$ T cells. That corresponds to a feeder cel ratio of 500-600:1. In Riddell Example 2, $75\text{-}90x10^6$ PBMC feeder cells and $15x10^6$ LCL feeder cells were used to expand $1.5\text{-}5x10^5$ T cells. That is a feeder cell ratio of 180-700:1. According to Ex. 3 and 4 the preferred ratio is 500:1 of PBMC feeder cells and 100:1 of LCL feeder cells, for a total of 600:1.

As disclosed in the specification at P9, L33-35, A 40 PD population (~ 10^{12} cells) has a mass of about 1kg. So $9x10^9$ cells (the highest expansion achieved by Riddell), is about 33 PD, and has a mass of about 10g T cells. With a feeder cell ratio of 600:1, a feeder cell mass of 6000g (6 kg) would be needed to practice Riddell's preferred REM method. To obtain that number of feeder cells would require 6000 blood donors each giving 1 liter of blood. This does not seem a reasonable expectation to us.

Assuming that Riddell nonetheless achieved a 1000-fold expansion of $9x10^9$ cells to $9x10^{12}$ cells (~43 PD), which is a cell mass of about 10 kg, to achieve the 50 PD population contemplated by **new** claim 89 (basis at P49, L32) by Riddell's method would require a feeder cell mass of 6000 kg.

That would call for 6,000,000 blood donors each giving 1 liter of blood, which is simply absurd.

The Examiner can readily calculate what Riddell would need in the way of blood donations to achieve the 60 PD population of

new claim 90 (basis at P9, L36 and P50, L30-34).

These calculations also assume that Riddell's REM method is just as efficient at expanding previously REM-expanded cells as it was in expanding "virgin" cells. Riddell has not established that is a viable assumption. It is not unusual in cell culture technology to find that repeated administration of a stimulant provides diminishing returns.

Neither Boel nor Santoli remedy the deficiency of the primary references.

Respectfully submitted,

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